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Engineering T cells for cancer therapy.

Mansoor W, Gilham DE, Thistlethwaite FC, Hawkins RE.

Cancer Research UK, Department of Medical Oncology, University of Manchester, Paterson Institute for Cancer Research, Christie Hospital NHS Trust, Withington, Manchester, UK.

It is generally accepted that the immune system plays an important role in controlling tumour development. However, the interplay between tumour and immune system is complex, as demonstrated by the fact that tumours can successfully establish and develop despite the presence of T cells in tumour. An improved understanding of how tumours evade T-cell surveillance, coupled with technical developments allowing the culture and manipulation of T cells, has driven the exploration of therapeutic strategies based on the adoptive transfer of tumour-specific T cells. The isolation, expansion and re-infusion of large numbers of tumour-specific T cells generated from tumour biopsies has been shown to be feasible. Indeed, impressive clinical responses have been documented in melanoma patients treated with these T cells. These studies and others demonstrate the potential of T cells for the adoptive therapy of cancer. However, the significant technical issues relating to the production of natural tumour-specific T cells suggest that the application of this approach is likely to be limited at the moment. With the advent of retroviral gene transfer technology, it has become possible to efficiently endow T cells with antigen-specific receptors. Using this strategy, it is potentially possible to generate large numbers of tumour reactive T cells rapidly. This review summarises the current gene therapy approaches in relation to the development of adoptive T-cell-based cancer treatments, as these methods now head towards testing in the clinical trial setting.

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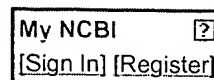
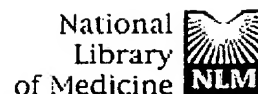
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Dendritic cells: therapeutic potentials.

Banchereau J.

Schering-Plough, Laboratory for Immunological Research, Dardilly, France.

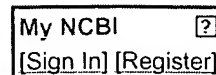
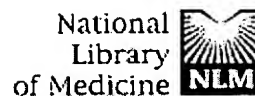
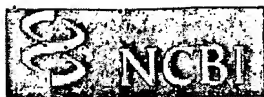
Dendritic cells (DCs) are leukocytes that are specialized to capture antigens and initiate T-cell-mediated immune responses. After capture of antigens, DCs, then in an immature stage, leave their tissue of residence and migrate through the lymph/blood into secondary lymphoid organs where they differentiate into mature cells. Because DCs can prime animals in the absence of any other adjuvant, they have been termed 'nature's adjuvant'. Large numbers of DCs can now be generated from circulating monocytes or from CD34 hematopoietic progenitors in response to GM-CSF in combination with either IL4 or TNF alpha. In mice, tumor antigen loaded DCs have been shown to prevent the development of tumors and even to induce the regression of established tumors. DCs therapy represents a very promising approach to the treatment of cancer and infectious diseases. Early studies indicate the existence of DC populations that can induce tolerance and may prove useful in organ transplantation.

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Functional improvement of damaged adult mouse muscle by implantation of primary myoblasts.

Irintchev A, Langer M, Zweyer M, Theisen R, Wernig A.

Department of Physiology, University of Bonn, Germany.

1. Myoblasts from expanded primary cultures were implanted into cryodamaged soleus muscles of adult BALB/c mice. One to four months later isometric tension recordings were performed in vitro, and the male donor cells implanted into female hosts were traced on histological sections using a Y-chromosome-specific probe. The muscles were either mildly or severely cryodamaged, which led to reductions in tetanic muscle force to 33% (n = 9 muscles, 9 animals) and 70% (n = 11) of normal, respectively. Reduced forces resulted from deficits in regeneration of muscle tissue as judged from the reduced desmin-positive cross-sectional areas (34 and 66% of control, respectively). 2. Implantation of 10(6) myogenic cells into severely cryodamaged muscles more than doubled muscle tetanic force (to 70% of normal, n = 14), as well as specific force (to 66% of normal). Absolute and relative amount of desmin-positive muscle cross-sectional areas were significantly increased indicating improved microarchitecture and less fibrosis. Newly formed muscle tissue was fully innervated since the tetanic forces resulting from direct and indirect (nerve-evoked) stimulation were equal. Endplates were found on numerous Y-positive muscle fibres. 3. As judged from their position under basal laminae of muscle fibres and the expression of M-cadherin, donor-derived cells contributed to the pool of satellite cells on small- and large-diameter muscle fibres. 4. Myoblast implantation after mild cryodamage and in undamaged muscles had little or no functional or structural effects; in both preparations only a few Y-positive muscle nuclei were detected. It is concluded that myoblasts from expanded primary cultures-unlike permanent cell lines-significantly contribute to muscle regeneration only when previous muscle damage is extensive and loss of host satellite cells is severe.

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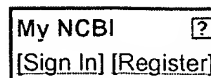
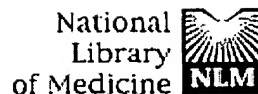
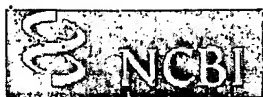
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Autologous chondrocyte transplantation for treating cartilage defects of the talus.

Baums MH, Heidrich G, Schultz W, Steckel H, Kahl E, Klinger HM.

Departments of Orthopaedic Surgery, Georg-August University Gottingen, Robert-Koch-Strasse 40, D-37075 Gottingen, Germany. mike.baums@freenet.de

BACKGROUND: Despite its highly specialized nature, articular cartilage has a poor reparative capability. Treatment of symptomatic osteochondral defects of the talus has been especially difficult until now. **METHODS:** We performed autologous chondrocyte transplantation in twelve patients with a focal deep cartilage lesion of the talus. There were seven female and five male patients with a mean age of 29.7 years. The mean size of the lesion was 2.3 cm(2). All patients were studied prospectively. Evaluation was performed with use of the Hannover ankle rating score, the American Orthopaedic Foot and Ankle Society (AOFAS) ankle-hindfoot score, a visual analogue scale for pain, and magnetic resonance imaging. **RESULTS:** All patients were available for follow-up at a mean of sixty-three months. There was a significant improvement in the Hannover score, from 40.4 points preoperatively to 85.5 points at the follow-up examination, with seven excellent results, four good results, and one satisfactory result. The AOFAS mean score was 88.4 points compared with 43.5 points preoperatively. Magnetic resonance imaging showed a nearly congruent joint surface in seven patients, discrete irregularities in four, and an incongruent surface in one. The patients who had been involved in competitive sports were able to return to their full activity level. **CONCLUSIONS:** The promising clinical results of this study suggest that autologous chondrocyte transplantation is an effective and safe way to treat symptomatic osteochondral defects of the talus in appropriately selected patients.

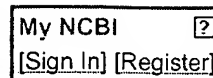
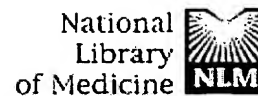
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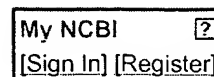
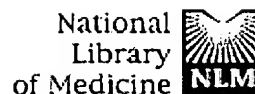
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Bone-cartilage transplantation from the ipsilateral knee for chondral lesions of the talus.

Baltzer AW, Arnold JP.

Center for Orthopaedic Surgery, Molecular Orthopaedics, and Neurosurgery, Dusseldorf, Germany. axel.baltzer@gmx.de

PURPOSE: We present a prospective analysis to review talus dome chondral and osteochondral lesions treated with autogenous bone-cartilage transplantation harvested from the ipsilateral knee since 1998. The clinical outcome of osteochondral defects is investigated by using a method for resurfacing that supplies hyaline cartilage. The outcome analysis also considers defect size and the number of transplanted osteochondral cylinders. **TYPE OF STUDY:** Prospective analysis of a case series. **METHODS:** Included in the study were 43 patients with ankle joint pain resulting from osteochondritis dissecans stage III-IV (n = 22), post-traumatic cartilage defects (n = 16), and focal osteoarthritis (n = 5). The mean age of this group was 31.2 years; there were 30 male and 13 female patients. To carry out the osteochondral resurfacing procedure, anteromedial or anterolateral arthrotomy (23 cases) or medial malleolar osteotomy (20 cases) of the distal tibia was performed. The osteochondral autograft transfer system (OATS; Arthrex, Naples, FL) was used for transplantation. The follow-up examinations were performed after 3 months (clinical, radiological), after 6 months (clinical, radiological), after 9 months (clinical, radiological, hardware removal, and second-look arthroscopy), after 12 months, and every following year (clinical, radiological, magnetic resonance imaging). The follow-up of 11 patients was greater than 2 years (maximum, 4.5 years), for 8 patients 1 to 2 years, for 12 patients 6 to 12 months, and for another 12 patients 0 to 6 months. The results have been validated by the scores of Evanski and Waugh score and Mazur et al. **RESULTS:** The mean pain intensity measured by visual analogue scale (0 to 10, with 10 representing the worst imaginable pain) reduced from 4.4 to 2.3 after 6 months (n = 34), to 1.6 after 1 year (n = 23), and after 2 years to 1.1 (n = 14). Patients reported a significantly improved range of motion of the ankle compared with their preoperative status. The smaller the diameter of the transplants and the smaller the number of transplants used, the better were the results in pain reduction and postoperative range of motion. The Evanski and Waugh score improved from 52 to 88 points and the score described by Mazur et al. from 53 to 90 of 100 possible points. All medial osteotomies were healed clinically and radiographically. All grafts showed bony integration in the



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[Mid-term results of autologous chondrocyte transplantation in knee and ankle. A one- to six-year follow-up study]

[Article in German]

Dorotka R, Kotz R, Trattnig S, Nehrer S.

Universitätsklinik für Orthopädie Wien, Wahringer Gürtel 18-20, 1090, Wien, Austria.

BACKGROUND: The reimplantation of autologous chondrocytes is a new technique in reconstruction of cartilage defects; initial results achieved with this technique have been promising. In an arthroscopic procedure, scales of cartilage are obtained from intact cartilage. The chondrocytes are then multiplied in special laboratories. A few weeks later, in a second procedure, the cartilage defect is filled with the cell suspension and closed with a flap of periosteum. **METHOD:** At our department, autologous chondrocyte transplantation (ACT) has been used in 10 patients since 1996, in 6 cases in the knee joint, and in 4 cases in the ankle joint. The mean age of the patients was 30 years. The mean size of the defect was 4 cm (2). In 4 patients, a parallel surgical procedure was required at the time of removal. **RESULTS:** The mean duration of follow-up was 21/2 years. Six patients had good to excellent results, 3 patients had moderate results, and one patient a poor result. The modified Cincinnati rating scale was improved from 2.4 to 7.1 points, and the Lysholm score from 59.2 to 86.6 points. The AOFAS score for ankle joints had improved from 33 to 76. **CONCLUSION:** We were able to show that ACT achieves improvement in the knee as well as ankle joint in the majority of patients. ACT appears to be a promising therapeutic concept for both joints.

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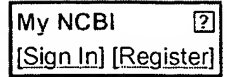
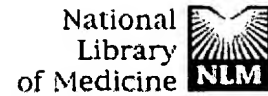
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Autologous chondrocyte implantation of the knee: multicenter experience and minimum 3-year follow-up.

Micheli LJ, Browne JE, Erggelet C, Fu F, Mandelbaum B, Moseley JB, Zurakowski D.

Division of Sports Medicine, Department of Orthopaedic Surgery, Children's Hospital, and Harvard Medical School, Boston, Massachusetts 02115, USA.

OBJECTIVE: To determine clinical outcome and graft survivorship in patients undergoing autologous chondrocyte implantation (ACI) for the repair of chondral defects of the knee. **DESIGN:** Prospective cohort study. **SETTING:** 19 centers in the United States. **PATIENTS:** 50 patients (37 males, 13 females). Mean age was 36 years (range: 19-53). Defects were grade III or IV with a mean size of 4.2 cm². All patients had a minimum of 36 months postoperative follow-up. **MAIN OUTCOME MEASUREMENTS:** Clinician and patient evaluation based on the modified Cincinnati Knee Rating System. Graft failure was defined as replacement or removal of the graft due to mechanical symptoms or pain. **RESULTS:** Clinician and patient evaluation indicated median improvements of 4 and 5 points, respectively, at 36 months following ACI ($p < 0.001$). Previous treatment with marrow stimulation techniques and size of defect did not impact the results with ACI. The most common adverse events reported were adhesions and arthrofibrosis and hypertrophic changes. Three patients had graft failure and required reimplantation or treatment with alternative cartilage repair techniques. Kaplan-Meier estimated freedom from graft failure was 94% at 36 months postoperatively (95% CI = 88-100%). **CONCLUSIONS:** These results of this study indicate excellent graft survivorship using ACI as well as substantial improvement in functional outcome.

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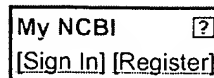
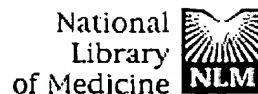
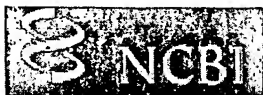
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Ex vivo gene therapy to produce bone using different cell types.

Musgrave DS, Bosch P, Lee JY, Pelinkovic D, Ghivizzani SC, Whalen J, Niyibizi C, Huard J.

Department of Orthopaedic Surgery, University of Pittsburgh, PA, USA.

Gene therapy and tissue engineering promise to revolutionize orthopaedic surgery. This study comprehensively compares five different cell types in ex vivo gene therapy to produce bone. The cell types include a bone marrow stromal cell line, primary muscle derived cells, primary bone marrow stromal cells, primary articular chondrocytes, and primary fibroblasts. After transduction by an adenovirus encoding for bone morphogenetic protein-2, all of the cell types were capable of secreting bone morphogenetic protein-2. However, the bone marrow stromal cell line and muscle derived cells showed more responsiveness to recombinant human bone morphogenetic protein-2 than did the other cell types. In vivo injection of each of the cell populations transduced to secrete bone morphogenetic protein-2 resulted in bone formation. Radiographic and histologic analyses corroborated the in vitro data regarding bone morphogenetic protein-2 secretion and cellular osteocompetence. This study showed the feasibility of using primary bone marrow stromal cells, primary muscle derived cells, primary articular chondrocytes, primary fibroblasts, and an osteogenesis imperfecta stromal cell line in ex vivo gene therapy to produce bone. The study also showed the advantages and disadvantages inherent in using each cell type.

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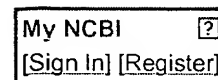
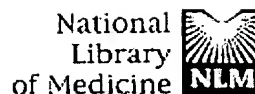
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Regulation of osteogenic proteins by chondrocytes.

Chubinskaya S, Kuettner KE.

Department of Biochemistry, Rush Medical College at Rush-Presbyterian-St. Luke's Medical Center, Chicago, IL 60612, USA. Schubins@rush.edu

The purpose of this review is to summarize the current scientific knowledge of bone morphogenetic proteins (BMPs) in adult articular cartilage. We specifically focus on adult cartilage, since one of the major potential applications of the members of the BMP family may be a repair of adult tissue after trauma and/or disease. After reviewing cartilage physiology and BMPs, we analyze the data on the role of recombinant BMPs as anabolic agents in tissue formation and restoration in different in vitro and in vivo models following with the endogenous expression of BMPs and factors that regulate their expression. We also discuss recent transgenic modifications of BMP genes and subsequent effect on cartilage matrix synthesis. We found that the most studied BMPs in adult articular cartilage are BMP-7 and BMP-2 as well as transforming growth factor-beta (TGF-beta). There are a number of contradicting reports for some of these growth factors, since different models, animals, doses, time points, culture conditions and devices were used. However, regardless of the experimental conditions, only BMP-7 or osteogenic protein-1 (OP-1) exhibits the most convincing effects. It is the only BMP studied thus far in adult cartilage that demonstrates strong anabolic activity in vitro and in vivo with and without serum. OP-1 stimulates the synthesis of the majority of cartilage extracellular matrix proteins in adult articular chondrocytes derived from different species and of different age. OP-1 counteracts the degenerative effect of numerous catabolic mediators; it is also expressed in adult human, bovine, rabbit and goat articular cartilage. This review reveals the importance of the exploration of the BMPs in the cartilage field and highlights their significance for clinical applications in the treatment of cartilage-related diseases.

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